1 539 625

PATENT SPECIFICATION (11) 1 539 625

(21) Application No. 27094/76 (22) Filed 29 June 1976
(31) Convention Application No. 7520456
(32) Filed 30 June 1975 in
(33) France (FR)
(44) Complete Specification published 31 Jan. 1979
(51) INT CL² B01J 13/00
(52) Index at acceptance
B8C A

(72) Inventors GUY VANLERBERGHE and ROSE-MARIE HANDJANI

(54) PROCESS FOR PREPARING AQUEOUS DISPERSIONS OF LIPID SPHERULES, AND AQUEOUS DISPERSIONS OF SUCH LIPID SPHERULES

(71) We, L'OREAL, a French Body Corporate, of 14, Rue Royale, Paris 8eme, France, do hereby declare the invention, for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement:—

It is known that certain lipids possess the property of forming, in the presence of water, mesomorphic phases, the state of order of which is intermediate between the crystalline state and the liquid state. Of those lipids which give rise to mesomorphic phases, some have already been stated to be able to swell in aqueous solution to form spherules dispersed in the aqueous medium, these spherules consisting of multi-molecular layers, preferably bi-molecular layers, having an approximate thickness of 30 to 100 Å (see, in particular, the article by Bangham, Standish and Watkins, J. Mol. Biol., 13, 238 (1965).

Hitherto it has only been possible to obtain lipid spherules consisting of concentric layers by using lipids containing an ionic hydrophilic group and a lipophilic group, and the processes of preparation which have been described generally produce spherules having a mean diameter of less than 1,000 Å. The process for obtaining these spherules consists of producing a dispersion, of which the disperse phase contains the lipid substance capable of forming the spherules, and of subjecting this dispersion to a treatment with ultrasonics; to produce the dispersion which is subjected to ultrasonics, it is possible first to produce, by evaporation on a wall surface, a thin film of the lipid substance to be dispersed, then (secondly) to bring the continuous phase of the dispersion which is to be produced into contact with the wall surface thus coated, and finally (thirdly) to agitate the system so as to obtain the dispersion which is to be subjected to ultrasonics. In another process, described in French Patent Application 2,221,122, it is possible, in order to obtain the dispersion to be subjected to ultrasonics, to add the lipid intended to form the walls of spherules to an aqueous phase and then to heat the mixture gently and agitate it vigorously by vibration. The spherules consisting of concentric layers which are thus obtained, and which have a maximum diameter of about 1,000 Å, are in general called liposomes.

It has already been proposed to utilise the liposomes to enclose aqueous solutions containing active substances, in the aqueous compartments contained between the double layers of lipid, and thus to protect the encapsulated substances from the exterior (see, in particular, the article by Sessa and Weismann, J. Lipid Res., 9, 310 (1968) and the article by Magee and Miller, Nature, Vol. 235 (1972)). Since the liposomes can have varying sizes below 1,000 Å, it is possible to vary their ability to penetrate into the human body, and this has made it possible to envisage numerous pharmaceutical uses, all the more so since their external electrical charge can make it possible to select the site where they will become fixed (Biochem J. (1971), 124 p. 58 P). However, as far as cosmetics are concerned, the use of spherules of diameter less than 1,000 Å can be disadvantageous because of the risk of penetration of the products through the skin. It is thus clear that at least for this type of application it would be desirable to be able to produce spherules with concentric lipid layers, which have a diameter greater than 1,000 Å.

Furthermore, the currently known processes for obtaining liposomes containing active substances between their concentric lipid layers suffer from considerable disadvantages. In the first place, the active substance introduced into the continuous phase of the dispersion which is subjected to ultrasonics, is only encapsulated to a very small extent between the lipid layers of the liposomes, be-

cause a very small part of the continuous phase of the dispersion finds itself trapped between the said layers. When it is desired to isolate the encapsulation liposomes it is necessary to pass the dispersion, which has been subjected to ultrasonics, over a separating column of the "Sephadex" (Registered Trade Mark) type, in which case the liposomes are obtained in the form of an extremely dilute dispersion. As a result, on the one hand, it is not possible in practice to obtain a high concentration of liposomes and, on the other hand, the active substance is only encapsulated to a very low degree and is lost when eluting the separating column without it being possible in practice to recover it in a simple manner, thus resulting in a large increase in the cost price of the active substances encapsulated in the liposomes. It is thus desirable to have available a process for the manufacture of spherules with concentric layers which makes it possible to obtain a dispersion having a high concentration of spherules with a consequent reduction in the loss of the product to be encapsulated between the layers of the spherules.

Finally, the processes of manufacture of liposomes which have been described hitherto mention that it is only possible to use certain well-defined categories of lipids: in the art quoted above, the use of phospholipids, of lipids containing an ionic hydrophilic group and a lipophilic group, and of unsaturated fatty acids has been mentioned.

The object of the present invention is to provide a process for obtaining an aqueous dispersion of spherules of diameter less than or greater than 1,000 Å at high concentrations, the said spherules being capable of encapsulating active substances, with a high encapsulation yield. In this specification, the word "encapsulate" is used to indicate that there is an aqueous phase present within a capsule consisting of lipid spherules. The process according to the invention can be applied to ionic or non-ionic lipids and thus makes it possible to include non-ionic lipid compounds amongst the lipids capable of forming spherules.

The present invention provides a process for preparing a dispersion of aqueous-liquid containing spherules consisting of ordered molecular layers enclosing an aqueous phase which is to be encapsulated, which comprises bringing into contact at least one liquid lipid which is dispersible in water and has the general formula:

X—Y

in which formula X represents an ionic or non-ionic hydrophilic group and Y represents a lipophilic group, with an aqueous phase to be encapsulated by the spherules, the lipophilic/hydrophilic balance of the lipid being such that the latter swells in the aqueous phase to be encapsulated, so as to form a lamellar phase, agitating the mixture to ensure mixing and obtain a lamellar phase, adding an aqueous liquid in an amount greater than the amount of lamellar phase obtained and agitating the resulting mixture vigorously for approximately from 15 minutes to 4 hours, generally 15 minutes to 3 hours to obtain the dispersion.

In a preferred embodiment, the weight ratio of the amount of aqueous phase to be encapsulated to the amount of lipids forming the lamellar phase is from 0.1 to 3; the aqueous phase to be encapsulated can be water or an aqueous solution of an active product; the weight ratio of the amount of dispersing liquid which is added to the amount of lamellar phase to be dispersed is 2 to 100; the dispersing liquid and the aqueous phase to be encapsulated are preferably isotonic (iso-osmotic); the dispersing liquid is advantageously an aqueous solution; the agitation carried out as the last stage of the process is achieved by means of a vibratory agitator; the process is carried out at ambient temperature or at a higher temperature if the lipid is solid at ambient temperature; where it is desired that the spherules obtained should have a mean diameter less than 1,000 Å, the dispersion of spherules can be subjected to a treatment with ultrasonics.

To form the lamellar phase, it is possible to use a single lipid or a mixture of lipids. The lipid or lipids, which are used, contains or contain a long lipophilic chain generally comprising from 12 to 30 carbon atoms, which can be saturated or unsaturated, branched or linear; in particular, it is possible to choose oleyl, lanolyl, tetradecyl, hexadecyl, isostearyl, lauryl or alkylphenyl chains. If the hydrophilic group of the lipid which forms the lamellar phase is a non-ionic group, it is possible to choose, as the hydrophilic group, for example, a polyoxyethylene, a polyglycerol an oxyethylenated or non-oxyethylenated polyol ester or, for example, a polyoxyethylenated sorbitol ester. If the hydrophilic group of the lipid which forms the lamellar phase is an ionic group, the hydrophilic group chosen is advantageously an amphoteric compound containing two lipophilic chains or an association compound of two long-chain organic ions of opposite signs. The term "long-chain" is intended to mean chains of at least 8, preferably 8 to 30 carbon atoms. Very satisfactory results have been obtained by using, as lipids which form the lamellar phase, polyglycerol ethers such as those which are described in French Patents Nos. 1,477,048 and 2,091,516 and in French Certificate of Addition 94,928.

The aqueous phase which is chosen for encapsulation can contain active substances of all kinds and in particular substances which

are of pharmaceutical or foodstuff value, or substances which are cosmetically active. The active substances can, for example, be the following, where cosmetics are concerned: products intended for the care of the skin and of the scalp, for example humectants, such as glycerol, sorbitol, pentaerythritol, inositol, pyrrolidone carboxylic acid and its salts; artificial bronzing agents, such as dihydroxyacetone, erythrulose, glyceraldehyde and γ-dialdehydes such as tartraldehyde (these products can optionally be associated with dyestuffs); water-soluble agents for protecting against sunburn; antiperspirants, deodorants, astringents, products having a freshening, tonic, cicatrising or keratolytic action, and depilatories; toilet waters; extracts of animal or vegetable tissues, such as proteins, polysaccharides and amniotic fluid; water-soluble dyestuffs, anti-dandruff agents, anti-seborrhoeic agents, oxidising agents (bleaching agents) such as hydrogen peroxide, and reducing agents such as thioglycollic acid and its salts. The following may be mentioned as pharmaceutical active substances: vitamins, hormones, enzymes (for example dismutase peroxide), vaccines, anti-inflammatory agents (for example hydrocortisone), antibiotics and bactericides.

It is obvious that lipids capable of stably encapsulating the aqueous phase will be chosen in accordance with the active substance contained in the aqueous phase to be encapsulated. In order that the lipids which constitute the lamellar phase should give stable spherules it is necessary that there should be sufficient lateral interaction between the lipid chains which, placed side by side, constitute the strata or layers of the spherules, that is to say that the Van der Waals forces between the chains should ensure sufficient cohesion of the layers. This conditions is generally satisfied in the case of the lipids having the characteristics given above. The lipids which can be used in the process according to the invention belong to the category of the emulsifiers of the water-in-oil type.

The process according to the invention makes it possible to obtain dispersions of spherules which consist of non-ionic lipid compounds and which therefore form new compositions which make it possible to encapsulate active substances which can be used, for example, in pharmacy, in nutrition or in cosmetics. The use of non-ionic compounds to form the encapsulating spherules is of not insignificant value where it is desired to avoid a situation where the spherules have an electrically charged external surface.

Accordingly, the present invention also provides a dispersion of spherules comprising ordered molecular layers of lipid compounds encapsulating an aqueous phase, the lipid compounds being non-ionic amphiphilic compounds having a lipid chain and capable of being dispersed in water, and the spherules having a diameter of 100 to 50,000 Å.

In a preferred embodiment, the spherules of the dispersion according to the invention contain an aqueous phase to be encapsulated; the non-ionic lipid compounds have a lipophilic/hydrophilic balance such that the compound swells in the aqueous phase to be encapsulated, forming a lamellar phase; the hydrophilic groups of the non-ionic lipid compounds are polyoxyethylenated groups, polyglycerolated groups, oxyethylenated or non - oxyethylenated polyol esters and, for example, polyoxyethylenated sorbitol esters; the non-ionic lipid compounds are preferably chosen from the group consisting of:

linear or branched polyglycerol ethers may have the formula:

$$R\text{—}(OCH_2CHOHCH_2\text{—})_n\text{—}OH$$

or

$$R\text{—}(\text{—}OCH_2\underset{\displaystyle CH_2\text{—}OH}{\underset{|}{CH}}\text{—})_n\text{—}OH$$

wherein n is an integer from 1 to 6, R is a linear or branched, saturated or unsaturated aliphatic chain of 12 to 30 carbon atoms, a hydrocarbon radical derived by the removal of —OH from a lanoline alcohol, or a 2 hydroxyalkyl radical derived from long-chain α-diols;

polyoxyethyleneated fatty alcohols;

oxyethylenated or non - oxyethyleneated polyol esters and in particular the esters of polyoxyethyleneated sorbitol; and

glycolipids of natural or synthetic origin, for example the cerebrosides.

The continuous phase of the dispersion, which surrounds the spherules, is an aqueous phase; the aqueous phase generally encapsulated in the spherules is an aqueous solution of active substance, preferably iso-tonic relative to the continuous phase of the dispersion.

Various additives can be associated with the non-ionic lipid compounds for the purpose of modifying the permeability or the surface charge of the spherules. In this context there may be mentioned the optional addition of long-chain alcohols and diols, of sterols, for example cholesterol, of long-chain amines and of their quaternary ammonium derivatives, of dihydroxyalkylamines, of polyoxyethyleneated fatty amines, of esters of long-chain amino-alcohols, of their salts and their quaternary ammonium derivatives, of phosphoric acid esters of fatty alcohols, for example sodium dicetyl-phosphate, of alkylsulphates, for example sodium cetyl-sulphate, and of certain polymers, such as the polypeptides and the proteins.

The present invention also provides a dispersion of spherules comprising ordered molecular layers encapsulating or enclosing an aqueous phase, these layers consisting of at

least one lipid compound of the formula XEY, wherein X denotes an ionic hydrophilic group and Y a lipophilic group, the liquid being dispersible in water and the spherules having a diameter of 1,000 Å to 50,000 Å.

In a preferred embodiment, the aqueous phase to be encapsulated is an aqueous solution of an active substance; the active substances of the aqueous phase to be encapsulated are products having a cosmetic action; the continuous phase, i.e. the liquid in which the spherules are dispersed of the dispersion is an aqueous phase; the ratio of the weight of the spherules relative to the weight of the continuous phase of the dispersion is from 0.01 to 0.5 and the continuous phase of the dispersion is advantageously iso-tonic relative to the aqueous phase encapsulated in the spherules.

The active substances, which can be encapsulated in the spherules of the two types of dispersion defined above, are extremely varied and correspond to those which have been indicated earlier for carrying out the process according to the invention. As a result, the compositions can be used in a variety of fields and in particular in pharmacy and in cosmetics.

The aqueous dispersions defined above are of very particular value in cosmetics because the use of large size spherules makes it possible to reduce the risk of these preparations passing through the skin.

It should be noted that the use of the aqueous dispersions according to the invention in cosmetics, whether dispersions containing non-ionic lipid compounds or dispersions containing ionic lipid compounds are concerned, offers a considerable advantage over the well-known use of emulsions. In fact, if it is desired to use preparations which simultaneously contain fatty substances and water, it is necessary, in order to ensure the stability of the emulsion, to use amphiphilic emulsifying compounds to ensure the stability of the dispersions. It is known that certain emulsifiers can have a relatively irritant effect when they are applied to the skin. It has been discovered, in the course of the work relating to the present invention, that this effect of the emulsifiers, for a given chemical structure, depends considerably on the form in which they are applied. Thus, it has been possible to demonstrate the fact that a water/oil emulsion composed of 42% of perhydrosqualene, 8% of an emulsifier and 50% of water is highly irritant, whilst an 8% strength aqueous dispersion of the same emulsifier has an irritation index which is practically negligible whilst perhydrosqualene is completely harmless. (Throughout this specification, percentages are by weight, unless otherwise stated. Accordingly, there is a synergistic irritation effect when an emulsifier and an oily phase are present. The aqueous dispersions according to the invention make it possible to avoid the simultaneous use of an emulsifier and an oil, which constitutes a considerable advance in the field of cosmetics.

It should be noted that it is possible to add to the dispersions of spherules according to the invention different auxiliary products, the object of which is to modify the presentation or organoleptic properties of the dispersions, such as opacifiers, gelling agents, aromas, perfumes or dyestuffs.

In general, the value of the dispersions according to the invention resides in the fact that they make it possible to introduce hydrophilic substances into an essentially lipophilic medium. As a result, under these conditions, the hydrophilic substances find themselves masked, resulting in protection against various possible agents which cause deterioration, such as oxidising agents, digestive juices and, more generally, compounds which are reactive towards the encapsulated substances. The penetration and/or fixing of the active substances can be varied by varying the size of the globules and their electrical charge. Their action can also be varied (producing a delay effect). Furthermore, the fact that they are masked makes it possible to suppress, or significantly to alter, their organoleptic properties, in particular the taste. Finally, the lipids used in these preparations in themselves possess a beneficial effect, for example an emollient effect, a lubricating effect, or the effect of glossiness.

In order that the object of the invention shall be better understood, several embodiments will now be described by way of purely illustrative and non-limiting examples.

EXAMPLE 1

500 mg of sorbitol trioleate oxyethyleneated with 20 mols of ethylene oxide (the product "Tween 85" marketed by Messrs. ICI Atlas, "Tween" is a Registered Trade Mark) are brought into contact with 0.335 ml of an 0.7 M aqueous solution of sorbitol in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

Thereafter 3 ml of a 1% strength aqueous solution of the product known by the Registered Trade Mark "Carbopol 934" (a polyacrylic acid crosslinked by polyallylsucrose, sold by Messrs. "Goodrich") are added. The flask is placed on a shaker and is agitated vigorously for one hour.

The dispersion obtained is gelled; the diameter of the spherules is greater than 1 micron.

EXAMPLE 2

250 mg of oleyl alcohol oxyethyleneated with 10 moles of ethylene oxides (the product "Brij 96" marketed by Messrs. ICI Atlas) and 250 mg of oleyl alcohol oxyethyleneated with

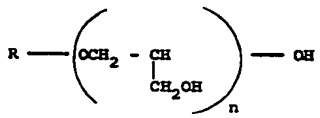
2 mols of ethylene oxide (the product "Brij 92" marketed by Messrs. ICI Atlas) are intimately mixed in a 50 ml round flask; the mixture obtained is then brought into contact with 1 ml of an 0.5 M aqueous solution of glycerol, and the whole is homogenised. The experiment is carried out at ambient temperature.

Thereafter, 20 ml of an 0.245 M solution of NaCl, are added. The flask, placed on a shaker, is agitated vigorously for 1 hour.

The dispersion obtained is fluid and milky; the diameter of the spherules is about 1 micron.

EXAMPLE 3

500 mg of the product of the general formula



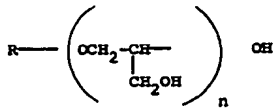
wherein R is the alkyl radical of hydrogenated lanoline alcohols and $\bar{n}$ has a statistical average value of 3, are brought into contact with 0.220 ml of a 0.5 M aqueous solution of pentaerythritol in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

4 ml of water are then added. The flask, placed on a shaker, is agitated vigorously for 30 minutes.

The dispersion obtained has a milky appearance; the diameter of the spherules is greater than 1 micron.

EXAMPLE 4

500 mg of the product of the general formula



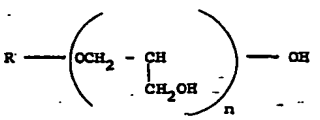
wherein R is the tetradecyl radical and n is equal to 2, are brought into contact with 0.75 ml of an 0.4 M aqueous solution of sorbitol in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at 40°C.

5 ml of water are then added. The flask, placed on a shaker, is agitated vigorously for 30 minutes.

The dispersion obtained is clear after treatment with ultrasonics; the diameter of the spherules is less than 1 micron.

EXAMPLE 5

500 mg of the product of the general formula



wherein R is the hexadecyl radical and n is equal to 2, are brought into contact with 0.335 ml of an 0.3 M aqueous solution of cysteine hydrochloride in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at 55°C.

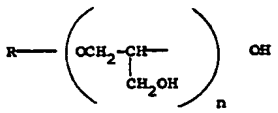
Thereafter, 4.1 ml of an 0.145 M solution of KCl are added. The flask, placed on a shaker, is agitated vigorously for 3 hours.

The dispersion obtained is practically limpid at 55°C; the diameter of the spherules is about 2 microns. On slowly cooling the dispersion to ambient temperature, a white opaque gel is obtained.

A sample of the dispersion taken at 55°C can be diluted with a solution, which may or may not be iso-osmotic, containing a thickener such as gums or polymers; a slightly opaque solution is obtained, the dilution ratio being chosen in accordance with the appearance of the solution which it is desired to obtain.

EXAMPLE 6

500 mg of the product of the general formula



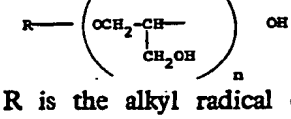
wherein R is the hexadecyl radical and n is equal to 2, are brought into contact with 10 ml of an 0.3 M aqueous solution of methionine in a 50 ml round flask placed in a waterbath at 55°C, and the mixture is homogenised. The experiment is carried out at 55°C.

The flask, placed on a shaker, is agitated vigorously for 3 hours at 55°C.

The dispersion obtained is limpid; the diameter of the spherules is about 1 micron. By cooling to ambient temperature, a white gel is obtained.

EXAMPLE 7

500 mg of the product of the general formula



wherein R is the alkyl radical of isostearyl alcohol and $\bar{n}$ has a statistical average value of 2, are brought into contact with 5 ml of water in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

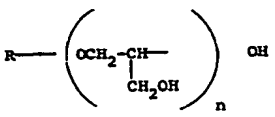
The flask, placed on a shaker, is agitated vigorously for 4 hours.

The dispersion obtained is milky; the diameter of the spherules is about 5 microns.

The dispersion can be subjected to ultrasonics in order significantly to reduce the size of the spherules.

EXAMPLE 8

83.2 mg 200 μmols) of the product of the general formula



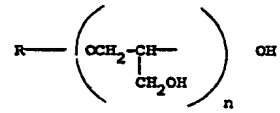
wherein R is the alkyl radical of oleyl alcohol and n is equal to 2, are dissolved in 2 ml of a mixture of chloroform and methanol (in the ratio of 2:1) in a 50 ml round flask. The solvent is evaporated by means of a rotary evaporator and the last traces of solvent are removed by exposure to the reduced pressure from a vane pump for one hour.

10 ml of an 0.3 M aqueous solution of glucose are brought into contact with the lipid. The flask, placed on a shaker, is agitated vigorously for 4 hours. The experiment is carried out at ambient temperature.

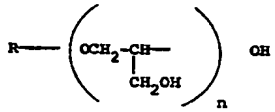
The dispersion is subjected to ultrasonics for 20 minutes so as to reduce the diameter of the spherules to a value of less than 0.5 micron. The dispersion is then filtered over a column of "Sephadex G 50 coarse" gel swollen in a 0.145 M solution of NaCl. The solution obtained is slightly bluish.

EXAMPLE 9

58 mg of the product of the general formula



wherein R is the alkyl radical of isostearyl alcohol and $\bar{n}$ is equal to 2, are intimately mixed with 58 mg of the product of the general formula



wherein R is the alkyl radical of "isostearyl" alcohol and $\bar{n}$ is equal to 6, in a 50 ml round flask, and the mixture obtained is brought into contact with 10 ml of a 1 M aqueous solution of glucose. The experiment is carried out at ambient temperature. The flask, placed on a shaker, is agitated vigorously for 4 hours.

The dispersion obtained is very fine; the spherules have a diameter of about 1 micron. The dispersion can be subjected to ultrasonics for 30 minutes so as to reduce the size of the spherules to a value of less than 0.5 micron.

The dispersions which contain either spherules of diameter greater than 1 micron or spherules of diameter less than 0.5 micron are filtered over a column of "Sephadex G 50 coarse" gel swollen in an 0.475 M solution of (NaCl, KCl).

EXAMPLE 10

500 mg of tetraethylene glycol monolauryl ether are brought into contact with 0.4 ml of an 0.3 M aqueous solution of glucose in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

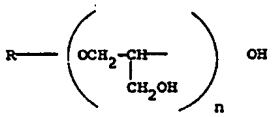
5 ml of an 0.145 M solution of KCl are then added.

The flask, placed on a shaker, is agitated vigorously for 15 minutes.

The dispersion obtained is limpid; the diameter of the spherules is about 1 micron.

EXAMPLE 11

500 mg of the product of the general formula



wherein R is the hexadecyl radical and $\bar{n}$ has a statistical average value of 3, are brought into contact with 0.5 ml of a solution containing 50 mg/ml of "Croteine C" (a protein of molecular weight about 10,000, marketed by Messrs. "Croda"), in a 50 ml round flask.

The mixture is homogenised. The experiment is carried out at 60°C.

4 ml of an 0.145 M solution of KCl are then added.

The flask, placed on a shaker, is agitated vigorously for 3 hours.

The dispersion obtained is limpid; the diameter of the spherules is about 1 micron. On slow cooling to ambient temperature, an opaque white gel is obtained.

EXAMPLE 12

300 mg of sphingomyeline are brought into contact with 0.350 ml of an 0.3 M aqueous solution of glucose in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

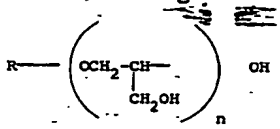
5 ml of an 0.145 M solution of NaCl are then added. The flask, placed on a shaker, is agitated vigorously for 2 hours.

The dispersion obtained is milky; the diameter of the spherules is about 2 microns.

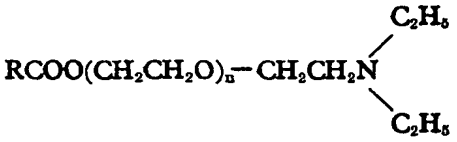
The dispersion can be subjected to ultrasonics for 1 hour so as to reduce the diameter of the spherules.

EXAMPLE 13

300 mg of the product, obtained by molecular distillation, of the general formula



wherein R is the alkyl radical of oleyl alcohol and n is equal to 2, 150 mg of cholesterol and 50 mg of an amine of the general formula



wherein RCOO is the copra radical and n is a number between 2 and 5, are intimately mixed in a 50 ml round flask and the mixture obtained is brought into contact with 0.5 ml of an 0.3 M aqueous solution of sorbitol; the whole mixture is then homogenised. The experiment is carried out at ambient temperature.

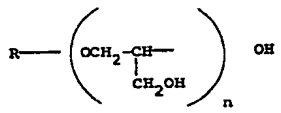
4 ml of an 0.145 M solution of KCl are then added.

The flask, placed on a shaker, is agitated vigorously for 4 hours.

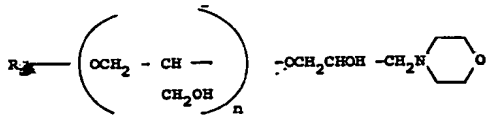
The dispersion obtained is opalescent; the diameter of the spherules is about 2 microns.

EXAMPLE 14

425 mg of the product of the general formula



wherein R is the alkyl radical of oleyl alcohol and n is a number equal to 2, and 75 mg of an amine of the following formula



wherein R is the oleyl radical and n has a statistical average value of 1, are brought into contact with 0.5 ml of an 0.3 M aqueous solution of glucose in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

4 ml of an 0.145 M solution of KCl are then added.

The flask, placed on a shaker, is agitated vigorously for 4 hours.

The dispersion obtained is opaque; the diameter of the spherules is greater than 2 microns.

The dispersion can be subjected to ultrasonics, whereupon the size of the spherules becomes less than one micron.

EXAMPLE 15

300 mg of sphingomyeline are brought into contact with 0.350 ml of an 0.3 M aqueous solution of ascorbic acid in a 50 ml round flask, and the mixture is homogenised. The experiment is carried out at ambient temperature.

2.650 ml of an 0.145 M solution of (NaCl, KCl) are then added. The flask, placed on a shaker, is agitated vigorously for 4 hours.

The dispersion obtained is milky; the diameter of the spherules is about 2 microns.

If desired, the dispersion can be filtered over a column of "Sephadex G 50 coarse" gel swollen in an 0.145 M solution of NaCl.

EXAMPLE 16

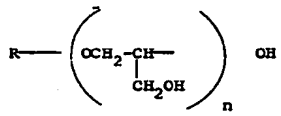
142 mg of the Na salt of N^2 - (tallow-alkyl) - N - dodecyl - N - (N,N' - diethyl-aminoethyl) - asparagine are dissolved in 2 ml of a mixture of chloroform and methanol (in the ratio of 2:1) in a 50 ml round flask. The solvent is evaporated by means of a rotary evaporator and the last traces of solvent are then removed by subjecting the product for one hour to the reduced pressure produced by a vane pump.

10 ml of an 0.3 M aqueous solution of glucose are brought into contact with the lipid.

The flask, placed on a shaker, is agitated vigorously for 4 hours. The experiment is carried out at ambient temperature. The size of the spherules is about 1 micron. The dispersion is then filtered over a column of "Sephadex G 50 coarse" gel swollen in an 0.145 M solution of KCl.

EXAMPLE 17

80 mg of the product of the general formula



wherein R is the hexadecyl radical and n is equal to 2, 10 mg of cholesterol and 10 mg of dicetyl phosphate are dissolved in 2 ml of a mixture of chloroform and methanol (in the ratio of 2:1) in a 50 ml flask.

The solvent is evaporated by means of a rotary evaporator and the last traces of solvent are removed by exposure to the reduced pressure from a vane pump for 1 hour.

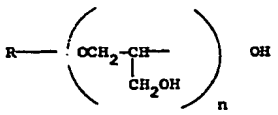
10 ml of an 0.15 M aqueous solution of the sodium salt of pyroglutamic acid are brought into contact with the lipids. The flask, placed on a shaker, is agitated vigorously for 2 hours on a waterbath at 55°C and is then cooled gradually until it has returned to ambient temperaature.

The dispersion is subjected to ultrasonics for 1 hour at a temperature which is kept at near ambient temperature. The dispersion is then filtered over a column of "Sephadex G 50 coarse" gel swollen in distilled water.

The dispersion obtained is fluid and clear after treatment with ultrasonics; the diameter of the spherules is less than 1 micron.

EXAMPLE 18

240 mg of the product of the general formula



wherein R is the alkyl radical of hydrogenated lanolin alcohols and n has a statistical average value of 3, and 60 mg of cholesterol are mixed intimately in a 50 ml round flask.

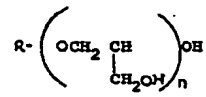
The mixture obtained is brought into contact with 0.4 ml of an 0.15 M aqueous solution of the sodium salt of pyroglutamic acid and the whole mixture is homogenised. The experiment is carried out at 45°C. 4.6 ml of a 9‰ solution of sodium chloride are then added.

The flask, placed on a waterbath, is agitated vigorously by means of a shaker for 2 hours at 45°C and then whilst cooling gradually until it has returned to ambient temperature.

The dispersion obtained is fluid and milky; the diameter of the spherules is greater than one micron.

EXAMPLE 19

200 mg of the product of the general formula



wherein R is the hexadecyl radical and n is equal to 2, 25 mg of cholesterol and 25 mg of dicetyl phosphate are intimately mixed in a 50 ml round flask; the mixture obtained is brought into contact with 0.3 ml of a 10% strength aqueous solution of tartraldehyde and the whole mixture is homogenised. The experiment is carried out at 55°C. 4.7 ml of an 0.145 M solution of NaCl are then added.

The flask, placed on a waterbath, is agitated vigorously by means of a shaker for 2 hours at 55°C and then whilst cooling gradually until it has returned to ambient temperature.

The dispersion obtained is gelled and has a slightly bluish appearance.

The simultaneous application to the skin of this dispersion of niosomes and of an aqueous solution, of the same final concentration, of tartraldehyde, makes it possible to assess two effects of the niosomes, which significantly intensify the colour developed and markedly improve the resistance of this colour to washing with water and with detergents.

WHAT WE CLAIM IS:—

1. Process for preparing a dispersion of aqueous liquid-containing spherules consisting of ordered molecular layers enclosing an aqueous phase which comprises bringing into contact at least one liquid lipid which is dispersible in water and has the general formula:

X—Y

in which X represents an ionic or non-ionic hydrophilic group and Y represents a lipophilic group, with an aqueous phase to be encapsulated by the spherules, the lipophilic/hydrophilic balance of the lipid being such that the latter swells in the aqueous phase to be encapsulated so as to form a lamellar phase, agitating the mixture thereby to obtain a lamellar phase, adding an aqueous liquid in an amount greater than the amount of the lamellar phase obtained and agitating the resulting mixture vigorously for approximately from 15 minutes to 4 hours to obtain the dispersion.

2. Process according to claim 1, in which the weight ratio of the amount of aqueous phase to be encapsulated to the amount of lipids forming the lamellar phase is from 0.1 to 3.

3. Process according to claim 1 or 2, in which the aqueous phase to be encapsulated is water.

4. Process according to claim 1 or 2, in which the aqueous phase to be encapsulated is an aqueous solution.

5. Process according to any one of claims 1 to 4, in which the weight ratio of the amount of liquid dispersing agent to the amount of lamellar phase is from 2 to 100.

6. Process according to any one of claims 1 to 5, in which the dispersing liquid and the aqueous phase to be encapsulated are isotonic.

7. Process according to any one of claims 1 to 7, which is carried out at a temperature at least equal to the melting point of the lipid.

8. Process according to any one of claims 1 to 7, in which the dispersion of spherules obtained is subjected to a treatment with ultrasonics such that the mean diameter of the spherules does not exceed 1000 Å.

9. Process according to any one of claims 1 to 8, in which the lipid contains a lipophilic chain having from 12 to 30 carbon atoms.

10. Process according to claim 9, in which the lipid contains an oleyl, lanolyl, tetradecyl, hexadecyl, isostearyl, lauryl or an alkylphenyl chain.

11. Process according to any one of claims 1 to 10, in which the hydrophilic group of the lipid is a non-ionic polyoxyethylene, polyglycerol or an oxyethylenated or non-oxyethylenated polyol ester group.

12. Process according to claim 11 in which the hydrophilic group is a polyoxyethylene.

13. Process according to any one of claims 1 to 10 in which the hydrophilic group of the

lipid is an amphoteric compound containing two lipophilic chains or an association of two long-chain organic ions of opposite signs.

14. Process according to any one of claims 1 to 8, in which the lipid is a linear or branched polyglycerol ether of formula:

$$R—(OCH_2CHOHCH_2—)_n—OH$$

or

$$R—(—OCH_2CH(CH_2—OH)—)_n—OH$$

wherein n is an integer from 1 to 6 and R is a linear or branched, saturated or unsaturated aliphatic chain comprising from 12 to 30 carbon atoms, a hydrocarbon radical derived by the removal of —OH from a lanolin alcohol, or a 2-hydroxyalkyl radical of a long-chain α-diol,

or a polyoxyethylenated fatty alcohol, an oxyethylenated or non-oxyethylenated polyol ester or a natural or synthetic glycolipid.

15. Process according to claim 14 in which the lipid is a polyoxyethylenated sorbitol ester or a cerebroside.

16. Process according to claim 11, 14 or 15 in which the non-ionic lipid is used in conjunction with a long-chain alcohol or diol, a sterol, a long-chain amine or a quaternary ammonium derivative thereof, a dihydroxyalkylamine, a polyoxyethylenated fatty amine, an ester of a long-chain aminoalcohol or a salt or quaternary ammonium derivative thereof, a phosphoric acid ester of a fatty alcohol, an alkyl-sulphate, a polypeptide or a protein.

17. Process according to claim 16 in which the non-ionic liquid is used in conjunction with cholesterol, sodium dicetyl-phosphate or sodium cetyl-sulphate.

18. Process according to any one of the preceding claims in which the aqueous phase to be encapsulated contains at least one cosmetic agent which is a humectant, an artificial bronzing agent, erythrulose, glyceraldehyde or an γ-dialdehyde, a water-soluble anti-sunburn agent, an antiperspirant, a deodorant, an astringent, a product having a freshening, tonic, cicatrising or keratolytic action, a depilatory, a toilet water, an extract of animal or vegetable tissues, a water-soluble dyestuff, an anti-dandruff agent, an anti-seborrhoeic agent, an oxidising agent or a reducing agent.

19. Process according to claim 18, in which the cosmetic agent is glycerol, sorbitol, pentaerythritol, inositol, a pyrrolidone, carboxylic acid or a salt thereof, dihydroxyacetone, a protein, polysaccharide or aminotic fluid, hydrogen peroxide or thioglycollic acid or a salt thereof.

20. Process according to any one of claims 1 to 17, in which the aqueous phase to be encapsulated contains at least one vitamin, hormone, enzyme, vaccine, anti-inflammatory agent, antibiotic or bactericide.

21. Process according to claim 20 in which the aqueous phase to be encapsulated contains dismutase peroxide or hydrocortisone.

22. Process according to any one of the preceding claims in which the dispersion contains at least one opacifier, gelling agent, aroma, perfume or dyestuff.

23. Process according to claim 1 substantially as hereinbefore described.

24. Process according to claim 1 substantially as described in any one of Examples 1 to 5 and 10 to 19.

25. A dispersion of aqueous liquid-containing spherules whenever prepared by a process as claimed in any one of the preceding claims.

26. A dispersion of spherules consisting essentially of ordered molecular layers enclosing an aqueous phase, said layer consisting of at least one lipid which is a non-ionic amphiphilic compound with a lipid chain, which can be dispersed in water, the spherules having a diameter of from 100 to 50,000 Å.

27. A dispersion according to claim 26 in which the lipid has a lipophilic/hydrophilic balance such that the compound swells in the aqueous phase thereby forming a lamellar phase.

28. A dispersion according to claim 26 or 27 in which the lipid is as defined in claim 11, 14 or 15.

29. A dispersion according to any one of claims 26 to 28 in which the lipid is present in conjunction with a substance as defined in claim 16 or 17.

30. A dispersion of spherules consisting essentially of ordered molecular layers enclosing an aqueous phase, said layers consisting of at least one liquid lipid of the formula X—Y, wherein X denotes an ionic hydrophilic group and Y denotes a lipophilic group the lipid being dispersible in water and the spherules having a diameter of from 1,000 Å to 50,000 Å.

31. A dispersion according to claim 30 in which the encapsulated aqueous phase is an aqueous solution of an active substance having a cosmetic action.

32. A dispersion according to claim 30 or 31, in which the ratio of the weight of the sepherules relative to the weight of the continuous phase of the dispersion is from 0.01 to 0.5.

33. A dispersion according to any one of claims 26 to 32, in which the continuous phase is an aqueous phase.

34. A dispersion according to any one of claims 26 to 33, in which the continuous phase is iso-osmotic relative to the encapsulated aqueous phase.

35. A dispersion according to any one of

claims 26 to 34 which has one or more of the features of the dispersion prepared by the process of any one of claims 16 to 22.

36. A dispersion according to claim 26 or 30 substantially as hereinbefore described.

J. A. KEMP & CO.,
Chartered Patent Agents,
14, South Square,
Gray's Inn,
London, WC1R 5EU.

Printed for Her Majesty's Stationery Office, by the Courier Press, Leamington Spa, 1979
Published by The Patent Office, 25 Southampton Buildings, London, WC2A 1AY, from which copies may be obtained.